Environmental Health Perspectives
Vol. 76, pp. 185-186, 1987

Issues in Biochemical Applications to Risk Assessment: How Do We Predict Toxicity of Complex Mixtures?

by Roy E. Albert*

Introduction

Predicting the toxicity of mixtures is an important current problem, and I'll talk about my personal view of the matter. I think that there are two broad classes of mixtures, one of which is the disposal mixtures: things that go into dump sites. These are highly variable from one site to another, since they depend on specific industrial operations, and it's difficult to extrapolate from site to site. The other class is the complex mixture from defined processes, such as combustion processes: automobile emissions, emissions from power plants, cigarette smoke, and so on.

I think the current approach to predicting the toxicity of complex mixtures is thoroughly empirical, and I don't know of any ways of doing things other than the way they're actually being done. Namely, with disposal mixtures, such as those occurring in dump sites, one summates the risk from individual agents for which one has existing data and ignores the possibility of interactions. It is recognized that this approach may be inadequate, but the problem of doing bioassays on individual dump sites is so expensive as to be daunting.

The situation which, in many respects, is easier is the case where the complex mixtures arise from defined processes of major economic importance, for example, the exhaust particulates from diesel engines. Here, it is economically feasible to mount large bioassay programs. I suspect that probably 20 million dollars has been spent on bioassays for diesel engine exhaust particulates by governmental and private sources. With mixtures like this, the approach is to identify the dominant effects, for example, with diesel particulates, cancer; cigarette smoke, the same; TCDD, the same, or reproductive effects.

The potency is quantitated for the mixtures using standard bioassays. Bioassays are also done to estimate the variability in the potency. For example, with diesel engines the carcinogenic potency can vary by a factor of 10, depending on the type of diesel engine that is used. Then the gravest and most sensitive effect is identified; that is, the effect which is thought to occur at the lowest level (for regulatory action). For example, TCDD is not only potent for carcinogenic effects, but also potent for reproductive effects. However, the acceptance of a low-dose linear nonthreshold response pattern for carcinogens has emphasized the carcinogenic effect of TCDD rather than the reproductive response.

So this is one person's view of how one goes about predicting the toxicity of complex mixtures, namely, that there are two approaches: the first is where one identifies the risk from the individual components for which there is data and accumulates the risk. And the other is where the mixture is treated as a single agent and potency variations in composition are taken into consideration (*1,2*).

And that ends my opening remarks. Anybody have comments or views to the contrary?

Discussion

DR. ERROL ZEIGER, NIEHS: I have one question with regard to the first approach, which is to identify the original components and then essentially sum across the risk. Has any work been done by summing across the individual risks of a mixture and coming up with the correct value?

DR. ALBERT: This really hasn't been looked into too much. Todd Thorslund at Clement Associates estimated the potency of about a dozen polycyclic aromatic hydrocarbons (PAHs) relative to benzo[a]pyrene from experiments in the literature where these agents were tested individually. He then estimated the potency of a PAH combination used by Schmahl in an experiment and found that there was a close correspondence between the estimated and observed potency of the combination of PAHs.

However, this still leaves open the issue of a complex mixture, such as cigarette smoke tar where the number of constituents can be well over 100,000, and where a large number of the constituents were unidentified. Here the question is whether the combined potency of

*Department of Environmental Health, University of Cincinnati Medical Center, Cincinnati, OH 45267.

a relatively few PAHs will simulate the potency of a highly complex mixture.

DR. BERNARD SCHWETZ, NIEHS: You're right that there's very little information on reconstructing the total toxicity of complex mixtures, and there's very little using carcinogenicity as an end point because of the cost. But if you look at the studies that have been done taking the most toxic components of complex mixtures and adding them for various end points of toxicity or the studies that have been done on simpler mixtures (e.g., binary, tertiary) and asking the question of whether additivity describes the toxic effects seen with the combinations, most often it's less than additive. And when surprises are seen where it's something beyond additive, it's seldom more than doubling in its effect.

So that the likelihood of seeing surprises in the form of synergistic interactions, where the data are totally unpredictive, is quite rare. Even when they are seen, they're fairly predictable by mechanism of action, for instance, the cholinesterase inhibitors.

DR. RAYMOND YANG, NIEHS: I have one comment and also I have something I'd like to sort of say in response to Errol Zeiger's question. The comment is I really admire your courage in taking this topic, because it seems to me from the literature that it's hard enough to predict with two as a combination when you deal with temporal relationships or with dose-response relationships, let alone complex mixtures.

With respect to Errol Zeiger's question, there are some studies in the literature. In fact, way back, maybe in the late sixties, Carol Weill and Henry Smith at the Mellon Institute did 27 industrial chemicals with two as the combination. They used what they called the harmonic formula of using $1/LD_{50}A + 1/LD_{50}2B$ to predict the toxicity. The paper was published in TAP [Toxicol. Appl. Pharmacol.]. There was also a Canadian group in the seventies that tested certain pairs of compounds which seemed to have synergistic effects.

We are initiating a program, the first stage of which is a study of 25 chemical mixtures in drinking water. The study is in contract negotiation. The second phase will be the selection of about eight chemicals that are most frequently seen in hazardous waste sites and groundwater contamination. We are looking for systematic experiments, with single chemicals, two as a group, three as a group, and so on. The test will most likely be a 14-day type of study with more end points than what's available in the literature.

These studies are in the planning stage. Hopefully in the future we can answer the questions discussed here a little better. Thank you.

DR. MARSHALL ANDERSON, NIEHS: Does it really surprise you that if you mix together a series of PAHs that they're additive because they're probably all working by the same mechanism? If I remember correctly, there is data indicating that mutations induced by the various polycyclics are proportional to the number of adducts. What I'm saying is that for the compounds of similar type you would expect they would be additive.

Now, suppose you had a mixture of things that acted differently. That would be your best bet for getting synergism. In the mouse skin, suppose you applied a mixture of something like TPA and of DMBA, each at low doses that wouldn't give tumors, then you would see tumor formation with the mixture. That's the kind of experiment that needs to be done to answer the question about mixtures instead of putting together things that you know are going to act the same way.

DR. ALBERT: Well, there's no question but there are concerns about interactions. The concern here is with the practical world. Ideally you do studies to characterize the behavior of the mixture as a whole. But more often than not, you're in no position to do so. You have to do the best by pulling out a few agents in the mixture and basing your overall estimate on what they add up to. It's not a very satisfactory situation.

REFERENCES

1. Environmental Protection Agency. Guidelines for the Health Risk Assessment of Chemical Mixtures. Federal Register 51: 34014–34025 (1986).
2. Albert, R. E., Lewtas, J., Nesnow, S., Thorslund, T. W., and Anderson, E. Comparative potency methods for cancer risk assessment applications to diesel particulate emissions. Risk Anal. 3: 101–117 (1983).